

Efficient Endoderm Induction from Human Pluripotent Stem Cells by Logically Directing Signals Controlling Lineage Bifurcations.

Journal: Cell Stem Cell

Publication Year: 2014

Authors: Kyle M Loh, Lay Teng Ang, Jingyao Zhang, Vibhor Kumar, Jasmin Ang, Jun Qiang Auyeong, Kian Leong Lee, Siew Hua Choo, Christina Y Y Lim, Massimo Nichane, Junru Tan, Monireh Soroush Noghabi, Lisa Azzola, Elizabeth S Ng, Jens Durruthy-Durruthy, Vittorio Sebastiano, Lorenz Poellinger, Andrew G Elefanty, Edouard G Stanley, Qingfeng Chen, Shyam Prabhakar, Irving L Weissman, Bing Lim

PubMed link: 24412311

Funding Grants: Antibody tools to deplete or isolate teratogenic, cardiac, and blood stem cells from hESCs

Public Summary:

Human embryonic stem cells (hESCs) can form all cell-types of the human body. In order to realize their therapeutic and regenerative potential, we must be able to precisely derive a pure population of a given human cell type (e.g., liver cells) from hESCs. This has been a challenging task to date, because hESC differentiation typically yields an impure population of several admixed cell-types, and contaminating unwanted lineages (e.g., bone) could potentially be hazardous if transplanted into human patients. Therefore there is a pressing need for methods to efficiently differentiate hESCs to desired cell-types in the absence of alternate fates. We met this challenge from a developmental biology perspective, namely by reexamining how various cell-types develop during embryogenesis. During embryogenesis, stem cells can frequently choose to differentiate into either a specific cell fate or another - and commitment to one occurs at the expense of the other (a so-called 'lineage bifurcation'). The principal conclusion of our study is that during hESC differentiation, in order to differentiate them down one lineage pathway (and not another), it was crucial not only to provide signals that positively drive the lineage of interest, but it was equally important to block signals that drive progression to alternate fates. Only then could stem cells be effectively differentiated into a pure population of a given fate in the absence of other lineages. Using the above principles, we could effectively differentiate hESCs into endoderm with extremely high (>99%) purity. Endoderm is the embryonic progenitor to various internal organs of therapeutic interest, including liver, pancreas, lungs and intestines. Being able to differentiate hESCs into very-pure endoderm populations, we could subsequently differentiate hESC-derived endoderm into downstream endodermal derivatives (including intestinal progenitors) with extremely high efficiency. We envision that these highly-pure endoderm populations will pave the way to generate many therapeutically-relevant cell types from hESCs and moreover the strategies we have conceived to logically direct lineage bifurcations could extend to other lineages such that we may be able to effectively guide hESCs down other lineage paths of interest.

Scientific Abstract:

Human pluripotent stem cell (hPSC) differentiation typically yields heterogeneous populations. Knowledge of signals controlling embryonic lineage bifurcations could efficiently yield desired cell types through exclusion of alternate fates. Therefore, we revisited signals driving induction and anterior-posterior patterning of definitive endoderm to generate a coherent roadmap for endoderm differentiation. With striking temporal dynamics, BMP and Wnt initially specified anterior primitive streak (progenitor to endoderm), yet, 24 hr later, suppressed endoderm and induced mesoderm. At lineage bifurcations, cross-repressive signals separated mutually exclusive fates; TGF-beta and BMP/MAPK respectively induced pancreas versus liver from endoderm by suppressing the alternate lineage. We systematically blockaded alternate fates throughout multiple consecutive bifurcations, thereby efficiently differentiating multiple hPSC lines exclusively into endoderm and its derivatives. Comprehensive transcriptional and chromatin mapping of highly pure endodermal populations revealed that endodermal enhancers existed in a surprising diversity of "pre-enhancer" states before activation, reflecting the establishment of a permissive chromatin landscape as a prelude to differentiation.